MORPHOLOGICAL STUDIES ON THE BEET ARMYWORM *SPODOPTERA EXIGUA* (HUBNER) (LEPIDOPTERA: NOCTUIDAE)

By

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To my wife, Ton-Nu Thanh-Chau

and my children,

Ngo Bao, Ngo Quynh, Ngo Anh, Ngo Anh-Thu.

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MORPHOLOGICAL STUDIES ON THE BEET ARMYWORM SPODOPTERA EXIGUA (HUBNER) (LEPIDOPTERA: NOCTUIDAE)

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The beet armyworm Spodoptera exigua (Hubner) (Lepidoptera: Noctuidae), a nearly cosmopolitan species, is an economically important pest in Florida and other areas. Morphological studies were concentrated on the antenna, wing scales, and the external genitalia and reproductive system of the adult, the egg, and first instar larva.

Female antennae are longer than male antennae. Six morphologically distinct types of sensilla were found on the antennae of each sex: Bohm bristles, chaetica, trichodea, auricillica, styloconica and coeloconica. However, only the male has the long sensilla trichodea. These are arranged in a distinct pattern. Wings can be divided into distinct zones based on types of scales:

two zones in forewing (dorsal), hindwing (dorsal and ventral), and three zones in forewing (ventral). Scent glands are present in the ventral intersegmental area between the 8th and 9th abdominal segments. Three types of sensilla are found on the lateral plates forming the ovipositor. The 9th abdominal segment of the male has a curved uncus and two lateral hooks arising from the claspers. Externally, there are many more scales and very few sensilla on the male genitalia compared to the female. The female ovary is composed of four ovarioles and development is panoistic.

The egg chorion around the micropyle is shaped petal-like with 7 - 11 lobes. Many tiny aeropyles occur on the edge of egg ridges.

The first instar larva is covered with microspines. Head setae tend to be sharp, curved and without raised pinaculum while body setae are straight and blunt tipped with pinaculum.

The haploid chromosome number for this species was determined to be n=31.

CHAPTER I

INTRODUCTION

The beet armyworm Spodoptera exigua (Hubner) is nearly cosmopolitan in distribution. Since first reported in Oregon (Harvey, 1876) it has gradually spread throughout the country reaching Florida in 1924 (Wilson, 1934). The larvae are very polyphagous and are important economic pests on many commercial crops grown in Florida including tomatoes, chrysanthemums, gladiolus, sweet corn, ferns, peppers and peanuts.

Difficulties in controlling the beet armyworm with insecticides along with concern for the environment have led entomologists to seek alternate methods of control for this and many other pest insects. These new methods of control require much more information on basic biology and morphology of insects. For example, the work on gametogenesis in the sugarcane borer Diatraea saccharalis (F.) by Virkki (1963) made possible the radiation and sterilization procedures for the male moths. The biology of the beet armyworm has been studied by Wilson (1932, 1934).

However, no detailed morphological studies of the insect have been done. The objective of this dissertation

is to describe the internal and external morphology of this insect with the aid of the scanning electron microscope with special emphasis on details of the reproductive and external sense organs.

CHAPTER II

LITERATURE REVIEW

Few detailed studies on the morphology and histology of the beet armyworm have been done. The morphology of the female sex pheromone glands of the beet armyworm was described along with 7 other noctuid species: Autographa californica (Speyer), Pseudoplusia includens (Walker), Rachiplusia ou (Guenee), Feltia subterranea (Fabricius), Heliothis zea (Boddie), Heliothis phloxiphaga (Grote), and Heliothis virescens (Fabricius), (Jefferson et al. 1966, 1968). The location of the glands of each species was described and some comparisons were presented. Hammad (1961) described scent glands of different noctuid species: Agrotis ypsilon (Hufnagel), Syngrapha circumflexa (Linnaeus), Earias insulana (Boisduval), Leucania loreyi (Duponchel), Pyrausta (Ostrinia) nubilalis (Hubner), Chilo simplex (Butler), Platyedra gossypiella (Saunders), and S. exigua. He reported S. exigua scent glands belonging to the brush-like type in the 8th abdominal segment.

The external morphology of the antennae of 4 noctuid species: Trichoplusia ni (Hubner), Heliothis zea, S. ornithogalli (Guenee), and S. exigua was described and different types of sensilla were classified but the

variations among the species were slight (Jefferson et al. 1970).

These are the only known studies on the beet armyworm, but there are reports on other species of Spodoptera. Most of these are listed under the generic name Prodenia which contained the majority of the species before being synonymized with Spodoptera.

Barth (1961) described the scent glands of S. ornithogalli and Hammad and Jarczyk (1958) reported on the morphology and histology of scent glands in S. littoralis [as P. litura] a closely related species occurring in Africa and the Middle East.

Many morphological studies have been done on S. litura (Fabricius), an economically important species occurring in India, Southeast Asia and some of the Pacific Islands. Various parts of the nervous system of the adult moths have been described by Srivastava (1967, 1970) and Mathur (1969). The larval nervous system has been described by Bahadur and Srivastava (1968a, 1968b) and Srivastava (1972). Srivastava and Mathur also described the morphology of the cephalic and prothoracic glands of the mature larva (1963) and the musculature of the head capsule of the mature larva (1964). Jefferson and Rubin (1970) clarified the description of the female sex pheromone gland and Mathur (1968) described the morphology of the excretory system of the larva. Abnormal variations in the wing venation were described by Bhattacherjee and Raghavan (1968).

The morphology and anatomy of the mature larva of S. littoralis [as Prodenia litura] was described by Hassan et al. (1958). Murad (1969) described the musculature of the sucking pump of S. mauritia (Boisduval). The male and female genitalia of S. mauritia acronyctoides (Guenee), S. pecten (Guenee), and S. abyssinia (Boisduval) were described by Chatterjee (1969).

Callahan's work on morphology and histology of Heliothis zea (1958), was particularly useful and many of his techniques were followed in this study. The morphology of the reproductive systems and mating in two other noctuids Pseudaletia unipuncta (Haworth), and Peridroma margaritosa (Hubner) were compared to Heliothis zea (Callahan and Chapin, 1960). Other detailed morphological descriptions of the reproductive systems of other Lepidoptera include those of Feltia subterranea (F.) (Snow and Callahan, 1968), Ephestia kuhniella (Zeller) (Norris, 1932), Pectinophora gossypiella (Saund) (Wellso and Adkisson, 1962), and Dioryctria abietella (D. & S.) (Fatzinger, 1970). Good histological descriptions have been given for those of H. zea (Callahan and Cascio, 1963) and E. kuhniella (Musgrave, 1937), and Choristoneura fumiferana (Clemens) (Outram, 1971).

Bullock and Horridge (1965) reviewed the types of sensilla occurring on insects. Research by Callahan (1969) and his associates (Callahan et al. 1968) described sensilla on the antenna of the corn earworm H. zea moth

as well as notes on some other noctuids. However, the only description of the sensilla on the beet armyworm antenna was made by Jefferson et al (1970).

The first microscopic study on the organization of the Lepidoptera wings was by Deschamps (1835). Bowerbank (1838) reported on the scale structure of the wings of Lepidoptera. Wonfor (1869) studied certain butterfly scales and gave some characteristics of sex. The taxonomic value of the scales of the Lepidoptera was reported by Kellogg (1894). Mayer (1896) gave further details on the development of the wing scales and their pigment in butterflies and moths. Kuznetsov (1915) contributed an overall study on Lepidoptera scale structure. Gentil (1935) studied in general the scale arrangement of the wings of Lepidoptera with an optical microscopy method. Since then, some researchers have studied some genera in more detail. The use of the transmission electron microscope on Morpho scales, (Gentil, 1942; Kinder and Suffert, 1943; Richards, 1944), showed that iridescent colors were structural and resulted from diffraction of light by ridges on the scale rather than from pigments. Yagi (1954) found that in the genus Colias (Lepidoptera: Pieridae) the yellow and/or orange colors were not structural and disclosed the presence of round and spindle-shaped aggregations of pigments. Kolyer et al (1970) studied the ultrastructure of the scales in Colias eurytheme and he found some variations

in color and position. This was the first time the scales were studied with the scanning electron microscope.

Research on the chromosome numbers of Lepidoptera has been done principally on butterflies, with very little work on moths in general and on the Noctuidae in particular. Among the 2,700 species of Noctuidae, the chromosome numbers of only 36 species have been reported (Robinson, 1971). The only member of the genus Spodoptera whose karyotype was determined (Gupta, 1964) was S. litura (as Prodenia litura).

CHAPTER III

METHODS AND MATERIALS

A. Rearing

The male and female moths of the beet armyworm used in this study were obtained from larvae reared on an artificial medium slightly modified (Bacheler and Emmel, 1974) from that used by Shorey and Hale (1965). All developmental stages were held at approximately 27° C and about 87% relative humidity with a 16 hour light and 8 hour dark day length. Larvae were reared in half-pint ice cream containers supplied with enough artificial food to complete larval development. Pupae were sexed and put into separate 1-pint ice cream containers except for those used to maintain the colony. An 8% sucrose solution was continuously available to the adults.

B. Gross Morphological Studies

For the reproductive system studies, two to three day-old male and female moths were dissected in physiological saline. The reproductive system was separated from the digestive tract, examined and photographed. Organs were preserved in alcohol for futher reference.

Histological studies of 4-day-old virgin moths were made of 3 parts; head-thorax, abdomen and wings. These parts were put into boiling Bouin's fluid, allowed to cool to room temperature for 2 days and then washed in 70% alcohol. Specimens were then imbedded in paraffin. Longitudinal and transverse sections were cut at 6 microns. Sections were stained with Mallory's triple stain method. Histological slides were observed with the Zeiss Photomicroscope II using Nomarski differential interference contrast. Photomicrographs were made of selected sections.

All measurements of wing scales were made with the aid of a micrometer disc in a compound microscope. All dimensions represent an average of measurements made on 12 scales. The measurements of internal reproductive organs were determined by measuring the structure on photographs with a millimeter rule photographed at the same microscope setting.

C. Chromosome Number

Chromosomes were studied in dividing cells in the testes of larvae of male moths. The testes were taken from the last larval stage and from the male adults 2 to 3 days after emergence. The testes were removed with No. 5 watchmaker forceps and placed on a standard microscope slide. The testes were then macerated and a few drops of lacto-aceto-orcein stain (Emmel, 1969) added. This preparation

was allowed to stand for about 20 minutes. The stained testes were next covered with a coverslip and the preparation was squashed first by thumb pressure between two pieces of blotting paper and then transferred to a laboratory press to exert uniform pressure and spread the preparation. The perimeter of the coverslip was then sealed with clear lacquer. Mounted slides were stored at 20° F for later reference.

Slides were inspected with a Zeiss Research Microscope Standard WL fitted with 25X and 40X plan apochromatic flat field objectives and automatic camera, and a Watson photo automatic phase contrast microscope "Hilux 70 PH" fitted with 40X planparo and 90X Fluorite contrast objectives and automatic camera. An oil immersion planapo 100X objective was used for critical observations.

D. Scanning Electron Microscope Studies

Eggs, first instar larva, antenna, wing scales, and the external reproductive organs of the adults were observed under the scanning electron microscope. Attention was primarily focused on sensory structures. The scanning electron microscope Cambridge Mark II-A was used for the microstructure of the insect through the courtesy of the USDA in Gainesville and with the aid of Mrs. Thelma C. Carlysle. Range of magnification used was x35 to x14,000.

Insect subjects were mounted on a stub with silver base paint and then introduced into a high vaccum evaporator. The high vaccum evaporator was a Denton DV-502 under a pressure of 2×10^{-5} TORR. Subjects were then coated with 200 angstrom thickness of gold before being transferred to the scanning electron microscope for observation. A camera (Tetronik Corporation 1:1) with polaroid film PN/55 4 x 5 was attached to the scanning electron microscope.

CHAPTER IV

RESULTS AND DISCUSSION

This research which is basically a descriptive research study is difficult to present in the standard dissertation format. For this reason, this section is subdivided into four sections: adult, egg, larva, and chromosome number.

A. Moth

The beet armyworm moth is about 2.2 cm across the wings (Figure 1, 2). The forewings are mottled grayish and the hindwings are off-white with dark scales along the veins and the border. Males (Figure 1) can usually be distinguished from females (Figure 2) by the more pointed abdomen tip.

1) Antennae

The antennae of the beet armyworm are setiform with a large number of flagellar segments referred to subsequently as the flagellum whereas the first two segments of the antennae are the scape and pedicel. There is considerable variation in number of segments from specimen to specimen, and even between antennae of a particular specimen. The mean number was 65 segments compared to 63 reported by

Jefferson et al. 1970. There was little difference in the mean number of segments between the sexes but there was some difference in the length and width of antennal segments between male and female (Table 1).

Table 1. Comparison of antennal flagellar segment length in beet armyworm (n=6 for each sex).

	Length (μ)		Width (μ)	
Females	mean	range	mean	range
Proximal segments	125.3	112-144	164.0	-114-180
Middle segments	132.8	127-139	100.0	91.108
Distal segments	88.8	84-91	63.2	57-72
Males				
Proximal segments	101.0	91-110	141.6	129-151
Middle segments	122.7	111-132	114.7	96-140
Distal segments	77.5	74-86	58.3	50-64

The basal segments of the antennae are short. The segments elongate distally, reaching their greatest length at about the midpoint of the antenna, then gradually decreasing in length to the end. The diameter of the segments decreases progressively from the base to the end of the antenna. All antennal segments of the male are shorter than the comparable ones of the females (Table 1).

The dorsal surface of the flagellum is covered by overlapping scales (Figure 3). Two rows of scales oriented

diagonally across each segment cover the dorsal surface of the antenna. Slightly more than half of the antennal circumference is covered with scales at the base of the antenna. Less of the antennal circumference is covered distally until only about one-third is covered with scales near the tip.

Most sensilla are found on the ventral surface of the flagellum (Figure 4, 5). At least six distinct kinds of sensilla were recognized. Some of these can be classified into subtypes. All dimensions given are means of 12 measurements (Table 2).

1) Bohm bristles (Figure 7, 8). The Bohm bristles are spine-like sensilla found only on the scape and pedicel. They taper more sharply than the sensilla chaetica of the flagellum. Two types of Bohm bristles occur; more numerous ones are short (type 1) and longer ones (type 2) which are evenly dispersed among type 1 have a socket and sclerotized ring at the base. Bohm bristles are longer in females, their average length varying from 24-39μ compared to 17-23μ in males.

On the scape, they are concentrated on 4 domes: two large domes laterally, and two small ones, one ventral and one dorsal. On the pedicel, they occur only on two lateral domes near the intersegmental area between the pedicel and the first flagellar segment.

2) Sensilla chaetica (Figure 6, 9). The sensilla chaetica are blunt spines set in a membraneous socket on the flagellar

segment. A scanning electron micrograph (at 6835X, Figure 11) shows that the surface of the spine is covered with radial ridges or striations.

Sensilla chaetica have a fixed position on both male and female antennae. Two chaetica are located on the scale side (dorsal) and are the only sensilla occurring there. One chaeticum is always located on each side of the ventral surface at the convergence point of the scale side with the sensory side. Sensilla chaetica are positioned at about the middle of the length of the segment. Their angle with the segment being about 35°. The basal segments have only one chaeticum on the sensory or ventral side. Two chaetica occur on the sensory or ventral surface at approximately the 20th segment making a total of six chaetica per segment.

The longest chaetica on both male and female moths are located on each side of the antenna at the convergence point of the scale side with the sensory side. The chaetica of the male are longer measuring approximately 108μ at the base of the antenna, reaching 132μ toward the middle and tapering to 94μ at the distal end. Chaetica on the female are 77μ at the base of the antenna, 91μ near the middle and tapering to 72μ at the tip.

Chaetica on the ventral surface of males measure from 77μ at the base of the antenna to 89μ near the middle and 79μ at the tip. The same chaetica on females are 43μ, 79μ, and 65μ respectively. The angle of chaetica on the ventral surface ranges from 45° to 80° on both male and female.

The chaetica on the scale or dorsal surface of the antenna are more consistent in length and make a smaller angle in relation to the antennal segment than any of the other chaetica. These chaetica range from 48μ long at the base of the antenna to 67μ at the tip. The angle is 15° to 35° in relation to an antennal segment. The chaetica originate at the distal end of the first row of scales.

3) Sensilla trichodea. The most numerous sensory hairs on the antennae can be separated into four types. a) Type 1: Consists of short porous trichodea having pores arranged in rows. The latter originate dorsally on the sensillum and continue a distally downward orientation to the ventral part of the sensillum (Figure 12). They measure 15-20μ in length and 1-2μ in diameter at base. Callahan (1969) called them stubby basiconica. b) Type 2: These are long porous trichodea measuring 20-30μ in length and 1-2μ in diameter at the base. The pore rows originate on the ventral part of the sensillum and extend on both sides of the sensillum in a dorso-distal direction until the two rows converge at the mid-dorsal point (Figure 13, 14). Both types of porous trichodea are thin-walled and are randomly arranged on the sensory side of the antenna. c) Type 3: These are nonporous trichodea (Figure 15). They are thick-walled and measure 38-58μ long and 2-5μ in diameter at the base. These sensilla are sharply pointed and arranged randomly over the entire sensory surface of both male and female antennae.

d) Type 4: These are extremely long trichodea found only on the male beet armyworm moth where they are arranged in six rows per segment with 5-6 trichodea per row measuring 84-125μ. Three rows originate ventro-laterally on each side of the antenna at the convergence point between the scale side and the sensory side. The long trichodea orient diagonally in rows.

All of the other sensilla types are located between the rows of long trichodea. Antennal segments bearing the long trichodea rows extend from the base of the antenna to approximately the 40th segment. Sensilla on the remaining segments are comparable to those on the female antenna. Ultrastructure of the nonporous trichodea (type 3) shows a fluted condition arranged circularly (Figure 15)

4) Sensilla coeloconica (Figure 16). These sensilla have been referred to as pit peg or picket-fence sensors (Callahan, 1969). Each sensillum consists of a porous peg with inward slanting spines oriented circularly around its periphery. Twelve to fifteen spines are arranged in a circular fashion with a fluted peg in the center. Spines curve inward toward the tip of the peg. Diameter of the pit is about 9-10μ on both male and female antenna. Pits occur randomly over the sensory surface of antenna with six to ten per segment. Significantly more sensilla were found in the 1st, 2nd, and 25th segments of females and on the 55th and penultimate segments of males (Jefferson et al. 1970).

5) Sensilla auricillica. This type of sensillum has been called spoon sensor (Griffith, 1968) and shoe-horn sensor (Callahan, 1969), however, sensilla auricillica as used by Jefferson et al. (1970) is more consistent with other latin terms. Sensilla auricillica are ear-like or spatula-shaped sensors averaging 12μ long and 5μ wide on both male and female. The wall is thin and porous with two to four occurring per segment. They are consistently located distally to the chaetica which are located at the convergence point of the sensory side with the scale side of antenna (figure 10, 17).

6) Sensilla styloconica (Figure 18). The sensilla styloconica are cone-shaped sensory structures mounted at the apices of stout cuticular pegs. Callahan (1969) called them taste rods. There is only one per segment, the size ranging from 16μ long on the basal segment to 29μ on the distal segment and 7.2μ in diameter. They occur at the distal end of each segment in the center of the sensory side.

The functions of all the sensors are not known, however electrophysiological research by Callahan (1968, 1969) and Jefferson et al (1970) indicate the probable function of some sensors (Table 2).

In both the male and female antennae, the mean total length is 2.1 times greater than the greatest mean width of any segments. The female antennae are longer than the male antennae. This would require that male antennae have more efficient detectors per unit length than the

female and consequently might indicate the reason for the multiple rows of trichodea on the male which are much longer than the regular ones, and which apparently serve as pheromone receptors. The great lack of neuroelectrophysiological information makes it impossible to assign reasons for morphological arrangements of the sensilla and consequently one can only conjecture about the placement of certain sensilla on the antennae.

Table 2. Measurements and proposed functions of sensilla on beet armyworm antenna.

Sensilla types	Proposed function	Length μ *	Diameter at base μ *
Sensilla coeloconica	chemoreceptor temp. & humidity	-	9.6
Sensilla auricillica	unknown	12	4.8
Sensilla styloconica	taste	15.6	7.2
Trichodea types 1, 2	pheromone receiver chemoreceptor	17-29	1-2
Trichodea type 3	pheromone receiver	38-58	2-5
Trichodea type 4 (♂only)	pheromone receiver	84-125	4-5
Chaetica ♂		55-123	5-10
Chaetica ♀		43-91	5-5.5
Bohm bristles ♀		24-39	
Bohm bristles ♂		17-23	

* n=12

Figure 1. Beet armyworm moth, male

Figure 2. Beet armyworm moth, female

Figure 3. Dorso-lateral view of antennal segment showing convergence of scale surface with sensory surface. Distal portion of antenna with dorsal surface to the right (X898)

Figure 4. Lateral view of the ventral surface of the basal portion of antenna (X705)

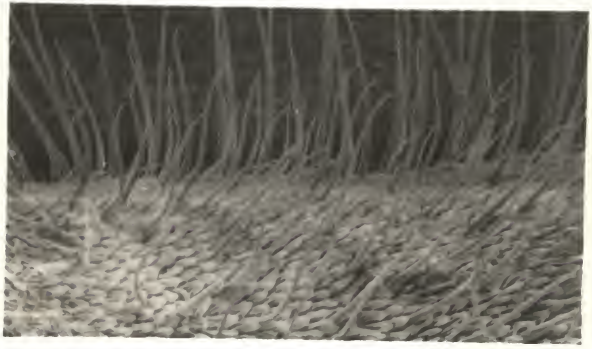
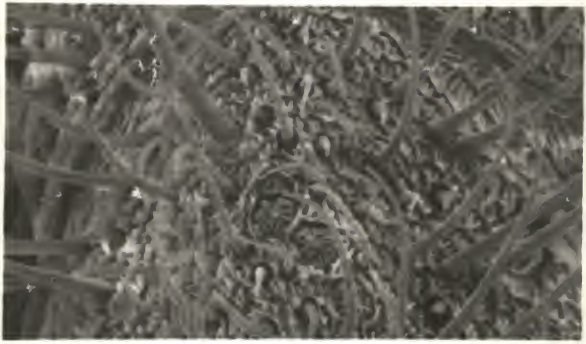


Figure 5. Most distal segment of the antenna showing sensors covering most of the circumference (X1410)

Figure 6. Chaeticum and trichodea on segment near base of antenna (X2926)



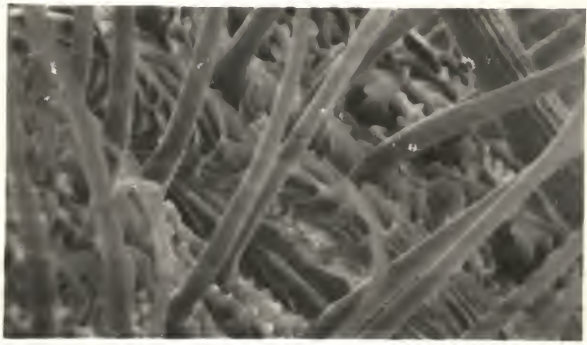


Figure 7. Bohm bristles on scape dome of male (X900)

Figure 8. Close-up of Bohm bristles on scape dome of female (X2667)

Figure 9. Close-up of chaeticum on the antenna (X7182)

Figure 10. Sensilla auricillica and button-like sensor (X7049)

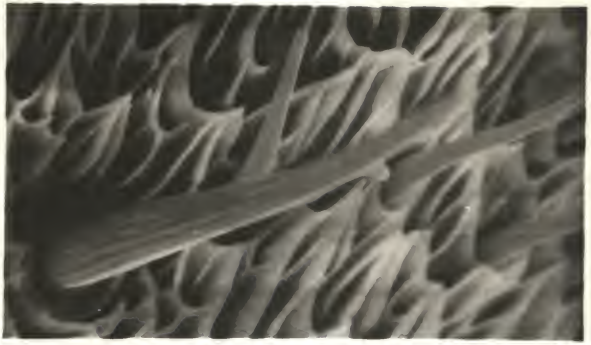


Figure 11. Close-up of chaeticum (X6835)

Figure 12. Short porous trichodea (X10640)

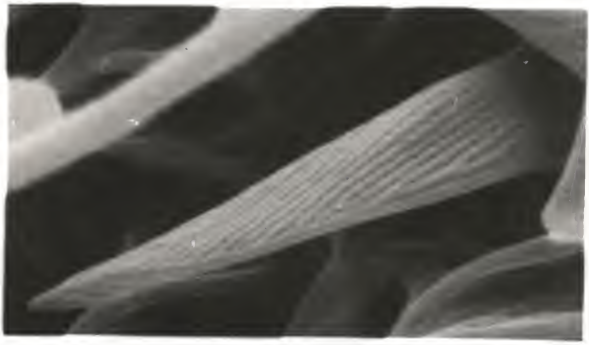
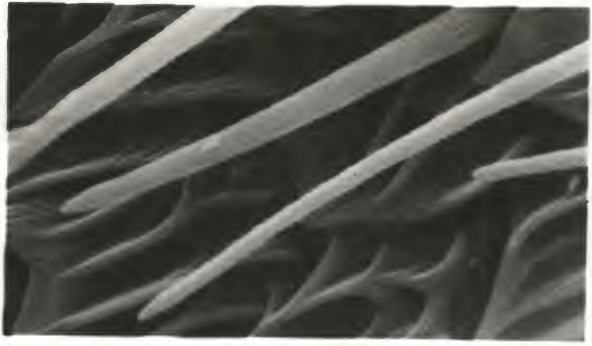


Figure 13. Long porous trichodea (X5060)

Figure 14. Long porous trichodea (X13,333)



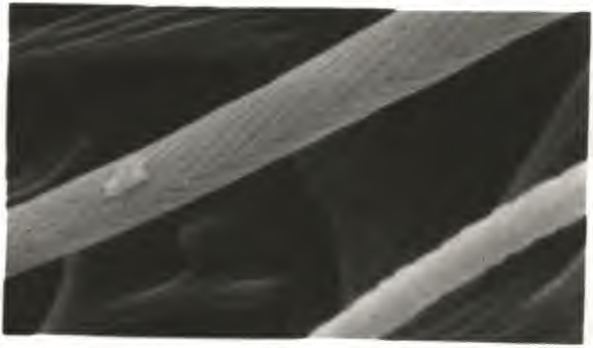
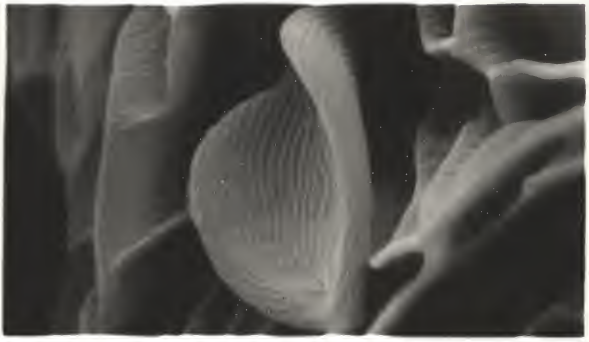


Figure 15. Non-porous trichodea (X6835)

Figure 16. Sensilla coeloconica (X6650)

Figure 17. Sensilla auricillica (X6835)

Figure 18. Sensilla styloconica (X4430)



2) Wings

Wings can be differentiated into certain zones based upon the types of scales found there. These zones do not correspond with wing venation. Approximate zones for both sexes and both wings (dorsal and ventral) are shown in Figures 19 and 20. In general the zones are similar on both wings dorsally and ventrally and in both sexes except for the ventral surface of the forewing which will be discussed later. Zone 1 usually takes up about the anterior one-fourth of each wing and zone 2 takes up the remainder of the wing. The third zone was the border consisting of 3 to 5 rows of scales.

Dorsal forewing: Scales found in zone 1 (Figure 21) of both male and female moths gradually increase in width from the shaft to the tip. They are serrate at the tip with 3 to 6 points in the males and 4 to 6 in the females. Average lengths are 292μ and 280μ in males and females, respectively. Average width is the same (41μ) for both sexes. Pigmentation is more heavily concentrated down the mid-line and the outer margin of the scale and across the apex. The scales in the female are darker than in the male.

There are two types of scales in zone 2. One type is similar to those in zone 1 in that they gradually increase in width from the base to the apex. The length and width of this type scale is 262 x 47μ for males and 276 x 56μ for females. Scales are serrate at the tip with 5 to 6

points for males and 5 to 7 for females. Heaviest pigmentation is concentrated along the mid-line from the shaft to the distal end of the scale and across the apex.

Interspersed under the serrate scales are the basal scales. These are blunt tipped scales 120 x 45μ (males) and 134 x 50μ (females). The blunt tip may be indented at 3 or 4 points in the females. There is less pigmentation in these scales than in the serrate scales (Figure 22).

The border scales are confined to 3 to 5 rows on the outer edge of the wing. These are elongate and the shaft comprises approximately half of the length. The width gradually increases from the shaft to the tip and both shaft and scale are pigmented. Scales are larger in males 610 x 78μ with 5 to 9 points compared to 470 x 53μ with 4 to 5 points in the female border scales. Width of the shaft is 7.2μ for males and only 5.0μ for females (Figure 23).

Ventral forewing: There are three zones in both male and female. Zone 1 takes up about one fifth of the wing area and zones 2 and 3 about two fifths each. The only difference between males and females is that zones 1 and 3 are adjacent at the base of the wing in the males while in the female, zone 2 completely separates zones 1 and 3 (Figure 20).

Scales found in zone 1 of both male and female moths gradually increase in width from the shaft to the tip. They are serrate at the tip with 4 to 6 points in the males and 3 to 5 points in the females. Average lengths are 312

and 251μ in males and females, respectively. Average widths are 56 and 44μ respectively (Figure 24).

There are two types of scales in zone 2. One type is serrate, similar to those in zone 1 with 6 to 7 points for males and 4 to 6 points for females. The length and width of this type scale is 280 x 50μ for males and 307 x 49μ for females. The second type consists of basal scales which are interspersed under the serrate scales. These scales are shorter and blunt at the tip (Figure 25), with a slight inward taper. Average measurements of these scales are 169 x 52μ and 140 x 51μ in males and females, respectively.

Covering scales in zone 3 are similar in both males and females. They are blunt tipped, but there are sometimes indentions in females (Figure 26). The ultrastructure is shown in Figure 28. Measurements are 315 x 55μ and 308 x 51μ in males and females, respectively. There are blunt basal scales interspersed under the covering scales in males. Their measurements are 96 x 48μ . There is no evidence of basal scales in females.

There is a sac-like structure (Figure 27) covered with spine-like scales at the posterior base of zone 3 (where the forewing and hindwing are joined). A sexual dimorphism was found with the scales smooth and larger at their base in females (Figure 29) while the scales are stronger and the diameter is about the same for the entire length in males (Figure 30). The function of the sac-like

structure is not known but it may minimize friction between the wings and facilitate the movements of both wings.

Dorsal hindwing: There are two zones in both males and females. Zone 1 covers the anterior one-fifth of the hindwing and zone 2 takes up the remainder. The anterior and posterior edges of the hindwing have many piliform (hair-like) scales varying from 600 to 1200μ long and 3.5 to 6μ wide. Only the males have these piliform scales interspersed in both zones 1 and 2, (Figure 31), there are none in the females in either zone. Covering scales are mostly light gray or white except the scales on the veins have darker pigmentation.

The covering scales in zone 1 are serrate with 2 to 5 points in males and 4 to 6 points in females. Pigmentation in zone 1 is darker than in zone 2. The measurements are 228 x 52μ for males and 252 x 58μ for females. The basal scales are blunt tipped and interspersed underneath the covering scales. Average measurements are 138 x 50μ .

The covering scales in zone 2 are serrate with 2 to 4 points for males and 4 to 6 points for females (Figure 32). Their average measurements are 220 x 55μ (males) and 238 x 53μ (females). White scales are predominant. There is no pigmentation in the females compared to a slightly darker pigmentation in the males. Blunt tipped basal scales are interspersed beneath the serrate scales. Their average measurements are 170 x 48μ .

The border scales are serrate with 3 to 5 points in the males and 4 to 5 points in the females. The shaft is one half

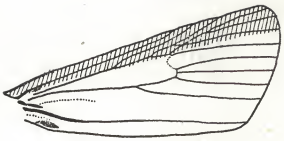
the total length of the scale. Their length varies from 438 to 860μ in males and 425 to 854μ in females. The average width is 40μ in males and 64μ in females. The scales are heavily pigmented. The width of the peduncle varies from 4.5 to 6μ. The most distinguishing characteristic to separate males from females is the frenulum spine number: there is only one in the males but there are three and sometimes four in the females.

Ventral hindwing: The zonal division is similar to the one of the dorsal surface. Covering scales in zone 1, are serrate with 4 to 5 points for both sexes (Figure 33). Their measurements are 252 x 48μ in males and 225 x 51μ in females. The scales are pigmented. The basal scales are blunt tipped and measure 101 x 49μ.

All of the scales are white in zone 2. The covering scales are serrate with 3 to 5 points in males and 3 to 4 points in females. Lengths of the scales are much shorter in females compared to those of males 125 to 240μ but the width is approximately the same, 50μ in males and 53μ in females. The basal scales are blunt tipped and interspersed beneath the covering scales. Their measurements are 100 x 51μ in males and 103 x 47μ in females (Figure 34).

There are four colors of scales: orange, white, gray and black with all variations between the last two. The color is not structural but due to pigment grains inside the scale. The white scales do not contain any pigment. There are no apparent differences at low magnification in form of different colored scales but the upper surface of

Figure 19. Fore- and hind-wing (dorsal) surface showing zonal division for both males and females (zone 1 shaded, zone 2 unshaded).



1
2

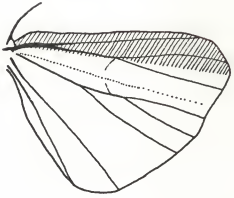


Figure 20. Upper: zonal division of forewing (ventral surface) in males

Middle: zonal division of forewing (ventral surface) in females

Lower: zonal division of hindwing (ventral surface) in both males and females

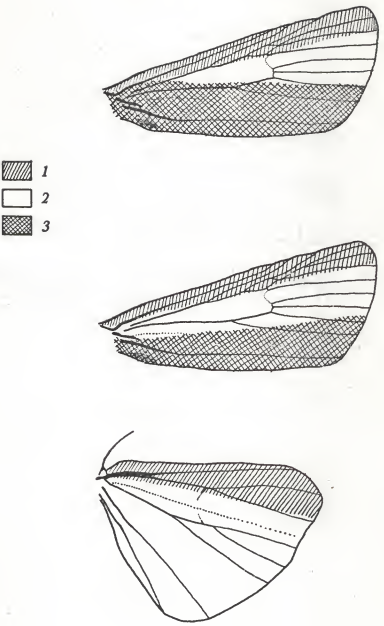

1
2
3

Figure 21. Dorsal forewing scales in zone 1 (X253)

Figure 22. Dorsal forewing covering and basal scales in zone 2 (X253)

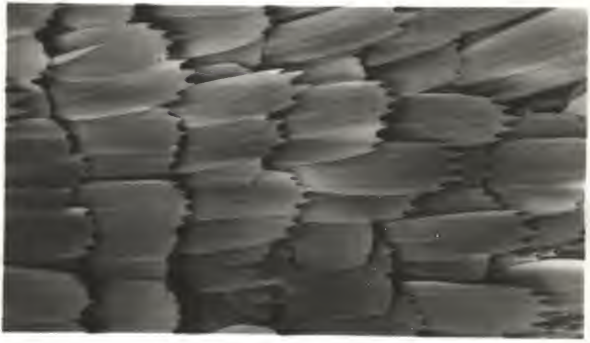


Figure 23. Border scales on dorsal forewing (X253)

Figure 24. Scales on ventral forewing in zone 1 (X253)

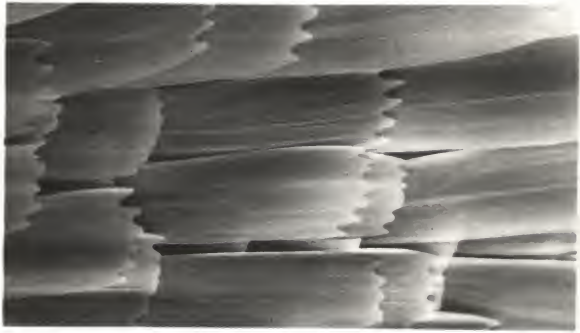
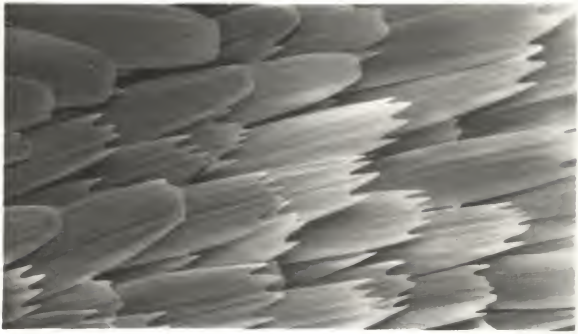


Figure 25. Basal scales and covering scales on ventral forewing in zone 2 (X253)

Figure 26. Scales on ventral forewing in zone 3 (X253)



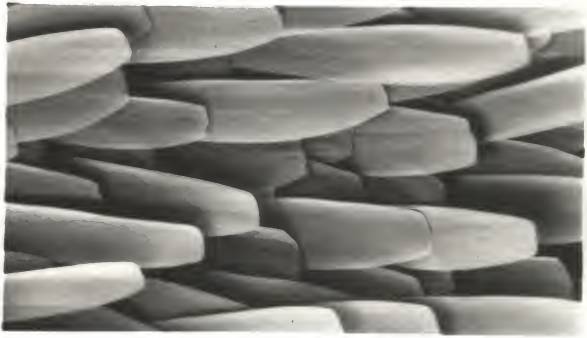


Figure 27. Spine-like scales on posterior base of ventral forewing on sac-like structure and regular scales in zone 3 (X226)

Figure 28. Ultrastructure of scales in zone 3 (X6317)

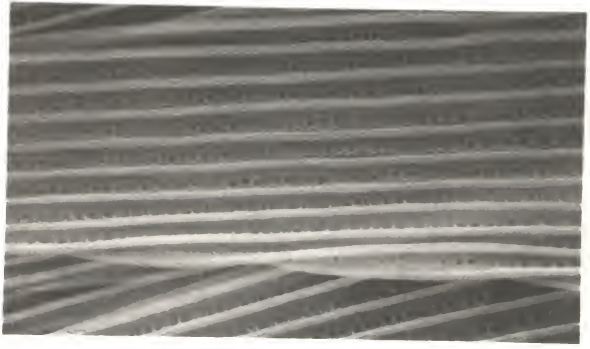


Figure 29. Scales on anterior anal area of the forewing (female, ventral) (X4522)

Figure 30. Scales on anterior anal area of the forewing (male, ventral) (X2660)

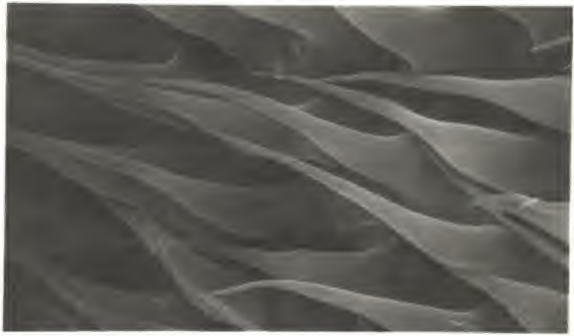
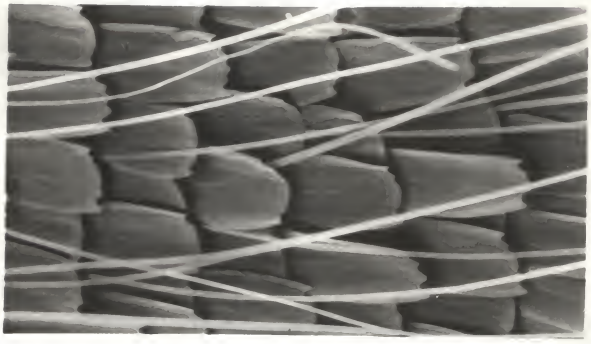


Figure 31. Dorsal hindwing: scales in zone 1 and piliform scales (X253)

Figure 32. Dorsal hindwing: scales in zone 2 (X253)



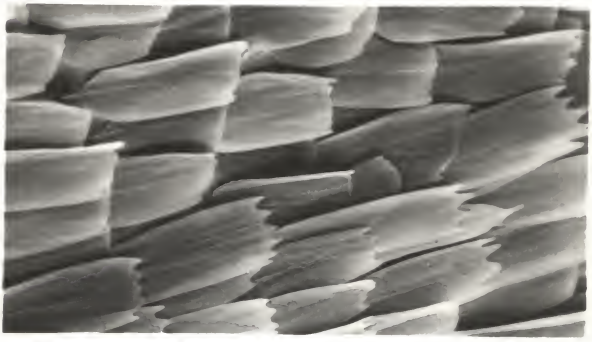
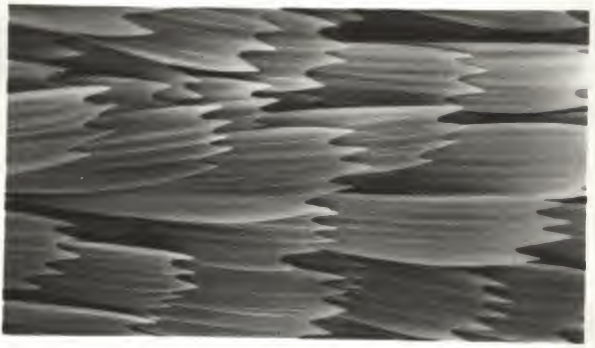


Figure 33. Ventral hindwing: scales in zone 1 (X253)

Figure 34. Ventral hindwing: scales in zone 2 (X253)



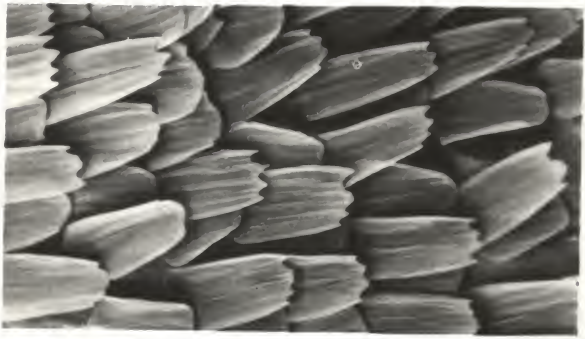


Figure 35. Ultrastructure of orange scale showing high longitudinal ribs and circular cross-ribs (X4985)

Figure 36. Ultrastructure of gray scale showing shallow grooves and irregularly patterned cross-ribs (X5852)

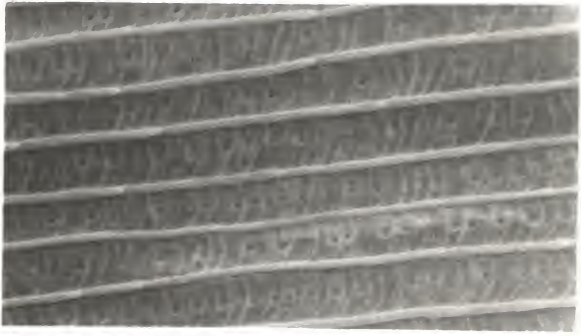
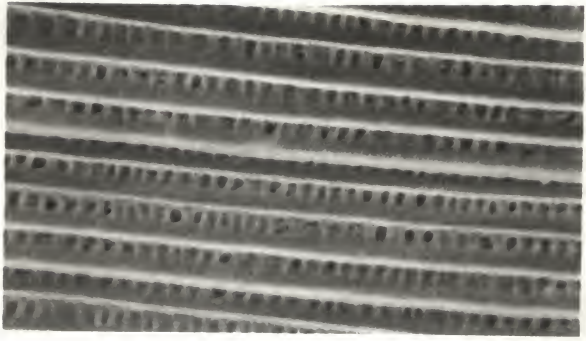


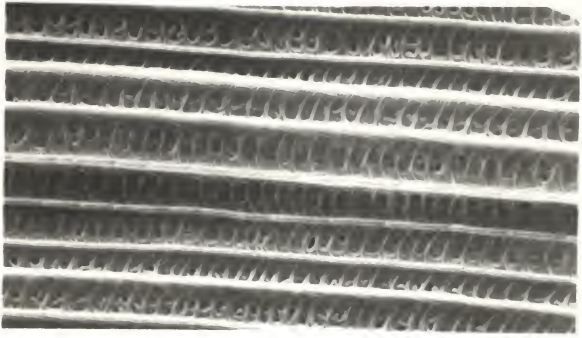
Figure 37. Ultrastructure of scale on ventral hindwing in zone 2 showing smooth groove without perforations (X4985)

Figure 38. Ultrastructure of border scale on ventral hindwing showing smooth groove with perforations (X5852)

Figure 39. Ultrastructure of the scale on dorsal forewing in zone 2 showing straight cross-ribs (X4985)

Figure 40. Ultrastructure of scale on dorsal hindwing in zone 2 showing curved cross-ribs (X4985)





scent ring (Jefferson et al, 1968). There was only one exception in the subfamily Amphipyrinae where the species Spodoptera litura has a glandular epithelium covering the entire 9th abdominal segment.

The scent glands are situated in the non-sclerotized area, between the 8th and 9th abdominal segments (Figure 41), contrary to Hammad's study on the beet armyworm (1961). A close-up picture of the scent gland (Figure 42) shows clearly glandular cells and the wavy spongy surface of the scent gland (Figure 43). There were different types of scales in the tip of the abdomen area but none of them are glandular scales as described by Hammad. There is no evidence of special secretory ducts in the epithelial cells. The pheromone is supposed to exude through the non-sclerotized area. The diameter of the scent glandular cells is about 10-15μ. The wavy surface of the gland area (microvilli) indicates that when the tip of the abdomen is extruded the glandular cells will expand fully through the largest surface and hence, provides the maximum release of pheromone. The location and structure of the scent gland of the beet armyworm is therefore similar to that of Spodoptera ornithogalli described by Barth (1961).

4) Reproductive System

a) External genitalia

The genital complex of Lepidopteran females includes the seventh through tenth abdominal segments. The eighth

the orange scales have higher longitudinal ribs and the cross ribs are consistently circular and accentuated when viewed at 4400X (Figure 35). The gray or black scales have relatively smooth, shallow grooves between the ribs and the cross ribs have no regular pattern (Figure 36).

Preliminary research on the ultrastructure of the scales at magnifications in excess of 4000X, revealed differences that offer possibilities for further classification of scale types. There are distinct differences in the structure of the ribs and cross ribs. The groove is smooth without perforations (Figure 37) or with perforations in some scales (Figure 38). The groove in others is perforated with numerous large openings (Figure 39). The cross ribs can be straight (Figure 39) or curved (Figure 40). Another variation is found in border scales on the forewing where every third or fourth rib is thickened. Much more could be done on the subject of ultrastructure of wing scales and a comparative study of various families of moths could reveal some very interesting and meaningful information on relationships throughout the order.

3) Scent Glands

Scent glands are situated in the ventral intersegmental area between the 8th and 9th abdominal segments. They belong to type 2 with ventral sac (Amphipyrinae) compared to type 1 with a dorsal sac or fold (Plusiinae) and type 3 with a

Figure 41. Scent glands located between VIIIth and IXth abdominal segments. Dark color: sclerotized area

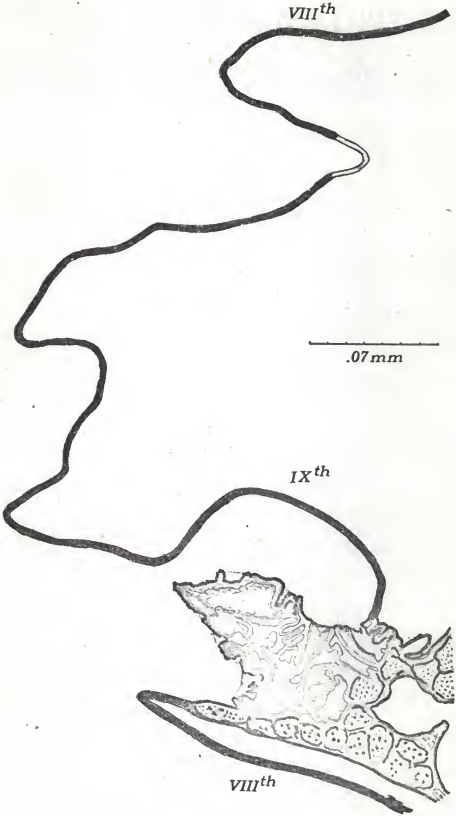

VIII th
.07 mm
IX th
VIII th

Figure 42. The scent gland in the intersegmental area between the 9th sternite (on the right) and the 8th sternite (on the left). Tip of abdomen at top of picture

Figure 43. Close-up showing wavy surface of the scent gland and glandular cells underneath. 8th abdominal sternite visible on the left. Tip of abdomen at top of picture

abdominal segment of S. exigua is modified with two lateral plates which serve as an ovipostor (Figure 44-46). These plates have three types of sensilla: trichodea, button-like, and sensory hairs (Figure 47). Scales are also observed on the inside of the plates (Figure 46).

The male genital complex of Lepidoptera includes the eighth, ninth, and tenth abdominal segment. The external male genitalia in the dorsal view (Figure 48) consists of the gnathos and 3 spine-like structures. The median spine-like structure, common to most Lepidoptera, is the uncus which articulates basally with the tegumen (a structure shaped like a hood or inverted trough lying dorsal of the anus). The paired spines articulate basally with the vinculum which is a U-shaped sclerite derived from the 9th abdominal sternite. Its arms articulate dorsally with the tegumen and its caudal margin with the harpes (or claspers). These two lateral hooks do not occur in some other genera of Noctuidae i.e. Feltia subterranea or Heliothis zea . These 3 spines apparently help the male position the female and hold her during copulation (Figure 48). The ventral view (Figure 49), shows the underside of the tegumen and uncus including the scale sockets on the tegumen. In Figure 50, the claspers form a V with the uncus between. The claspers are thickly clothed with scales which may have a sensory function (Figure 51).

Thus the external genitalia of the male consists of the vinculum which lies ventral to the aedeagus. The left and right arms are actually a single piece forming the sternum of the ninth abdominal segment. The tergal area of the ninth segment, called the tegumen, is fused with the coxosternal arc, the vinculum, to form the base (Figure 49) of the male genitalia. The paired claspers are appendages of the 9th segment and articulate at the coxopodite area of the vinculum. The uncus and aedeagus are structures of the tenth segment. Some of the same sensilla that are found on the antennae are found on the male genitalia: such as chaeticum (Figure 52), and trichodea. The ultrastructure of the scales (Figure 53) shows similar longitudinal ridges and transverse veins as found in the wing scales but without the holes.

The striking difference between the male and female genitalia is that there are many more scales and very few sensilla around the male genitalia compared to the female. Why sensors are much more numerous in the female genitalia is not known, however, it is likely associated with the necessity of the female to locate a suitable oviposition site using the ovipositor as a test organ.

b) Reproductive organs

The major morphological difference between reproductive systems of noctuid species are found in the bursa copulatrix and "ovipositor" of the female and in the

claspers or harpes, aedeagus and cuticular portion of the ductus ejaculatorius simplex of the male.

The female reproductive system consists of a pair of ovaries which open to a pair of lateral oviducts (Figure 54). Each ovary is composed of 4 ovarioles (or egg tubes). Ovariole development of the oocytes is panoistic. The maturity of the eggs is indicated by the color: white at the beginning, then pink-purple, and finally greenish when the eggs are mature. The two lateral oviducts join the median oviduct which opens to the gonopore (oviporus and anus) at the 9th segment. Two other glands open to the gonopore:

1) The paired accessory glands consist of small and long tubules which are enlarged at the base to function as reservoirs of the accessory glands and open to a median duct joining the gonopore.

2) The spermatheca also opens to the gonopore. It is composed ot three parts:
   a) large lobe (utriculus) of spermatheca
   b) upper loop of ductus receptaculi (spermathecal duct)
   c) convoluted area and lower loop of ductus receptaculi.

There are two ducts at the base of the spermatheca, one opening to the gonopore and the other leading to the base of the bursa copulatrix.

The gonopore which consists of oviporus and anus is on the ninth abdominal segment while the copulatory vulva opens on the eighth abdominal segment.

The bursa copulatrix is the most conspicuous organ in the female insect. The sides are twisted and ridged with sclerotized ribs. At the top of the sac-like organ is the place where the capsules of the spermatophores are formed. The number of spermatophores indicates the number of copulation times. The most spermatophores found in the bursa copulatrix of any moth was four. They accumulate one after another. Williams (1948) listed three classes of Lepidoptera using spermatophores as criteria: class A, having the aperture of the spermatophore in direct contact with the seminal duct, of which the beet armyworm is an example, class B, having a large secretion-filled reservoir between the aperture of the spermatophore and the seminal duct and class C, which includes a few primitive families that have no seminal duct and only one external opening instead of two as is typical of most Lepidoptera. The spermatophore is composed of a capsule and a sclerotized tube or collum (Figure 55). The neck-connection between the capsule and the collum is elbowed making the capsule vertical to the tube. The capsule loses its spheric shape and becomes flattened when the sperm are released through the seminal duct but the collum remains a hard sclerotized structure during the life time of the female.

The adult male reproductive system has only one fused testis as compared to two separate testes in the last larval instar. The two large bean-like testes in the

Table 3. Measurements of female internal reproductive organs and spermatophore.

Organs		Mean length (in mm)*
Accessory glands		5.5
Reservoir of accessory glands		.8
Spermatheca (utriculus)		1.2
Spermathecal duct		1.6
Lower loop of ductus receptaculi		.6
Bursa copulatrix		3.0 length, 1.2 width
Ovarioles		29.1
Vaginal duct		1.5
Seminal duct		1.9
Oviductus communis		2.5
Lateral oviduct		1.8
Spermatophore	(Length)	(Width)
Capsule	1.2-2.0	1.0-1.3
Neck	1.4-2.0	.2-.3(diameter)
Collum	3.0-4.0	.2-.3(diameter)

*n=6

larval stage shrink and fuse in the prepupal stage as the larva shrinks and decreases in size. The moth testis is located around the fourth and fifth abdominal segments. It is spherical and rose-colored. The paired seminal vesicles lead from the testis to the vas deferens which

open into the ductus ejaculatorius duplex near the midpoint (Figure 56). The seminal vesicles are about three-times as wide as the vas deferens. Both are creamy-white like the remaining glands and ducts of the organs. The two accessory glands-as two parallel ducts-are joined together their entire length before they open into the basal end of the ductus ejaculatorius duplex. The latter fuses into the ductus ejaculatorius simplex which is the longest duct of the reproductive organ. About one-sixth of the distance from the beginning, there is an enlargement of the duct. The ductus ejaculatorius simplex leads to the endophallus. The aedeagus is the sclerotized part surrounding the endophallus.

Table 4. Measurements of male internal reproductive organs.

Organs	Mean length (in mm)*
Seminal vesicles	1.5
Vas deferens	2.0
Accessory glands	24.0
Ductus ejaculatorius duplex	6.1
Ductus ejaculatorius simplex	51.2
Testis	1.2 (in diameter)

*n=6

Figure 44. Ovipositor showing sensilla on extreme tip (X133)

Figure 45. Close-up of ovipositor opening (X253)

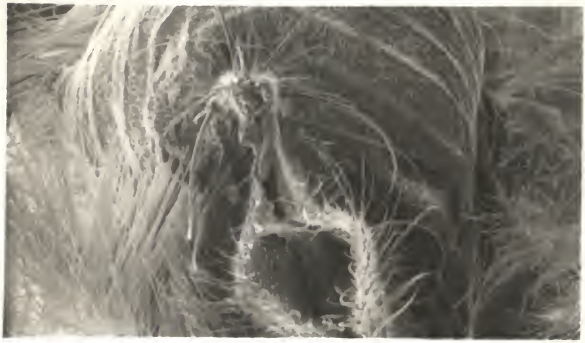


Figure 46. Close-up of ovipositor (X525)

Figure 47. Sensilla on dorsal posterior ovipositor (X1330)

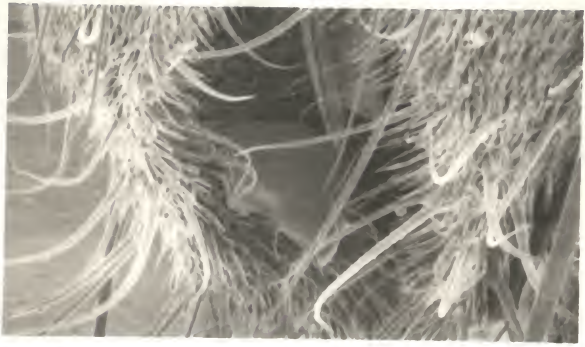


Figure 48. Dorsal view of male genitalia showing gnathos, uncus, and spines of claspers (X665)

Figure 49. Male tegumen supporting uncus with claspers on either side (X825)

Figure 50. Dorsal view showing uncus and claspers (X66)

Figure 51. Inside surface of claspers of male genitalia (X452)

Figure 52. Sensilla and scales on outside of the clasper; male genitalia (X1463)

Figure 53. Ultrastructure of scale inside of clasper; male genitalia (X7182)

Figure 54. The female reproductive system:

A. bursa copulatrix
B. vaginal duct
C. vulva
D. ovaries
E. lateral oviduct
F. median oviduct
G. oviporus and anus (gonopore)
H. spermatheca (utriculus)
I. spermathecal duct
J. lower loop of ductus receptaculi
K. accessory gland
L. reservoir of accessory gland

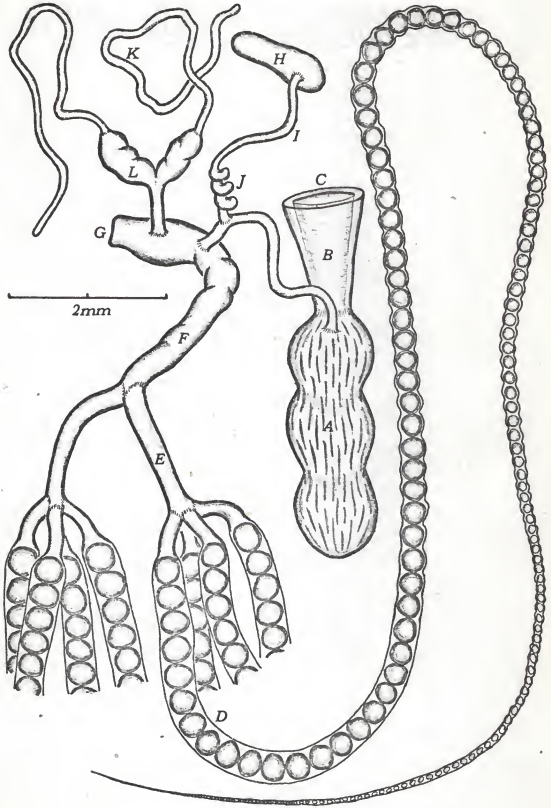

K
H
I
L
J
C
G
B
2mm
F
A
E
D

Figure 55. The spermatophore:

A. capsule
B. neck
C. collum
D. sperm bundles

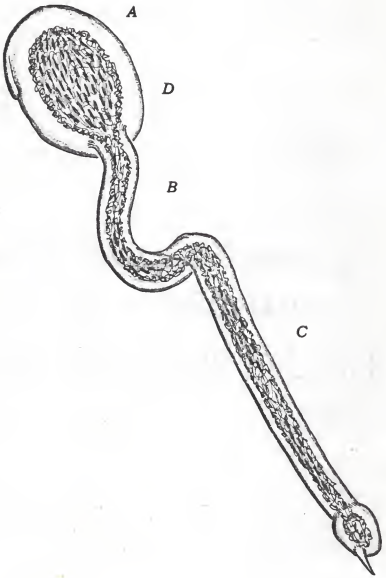
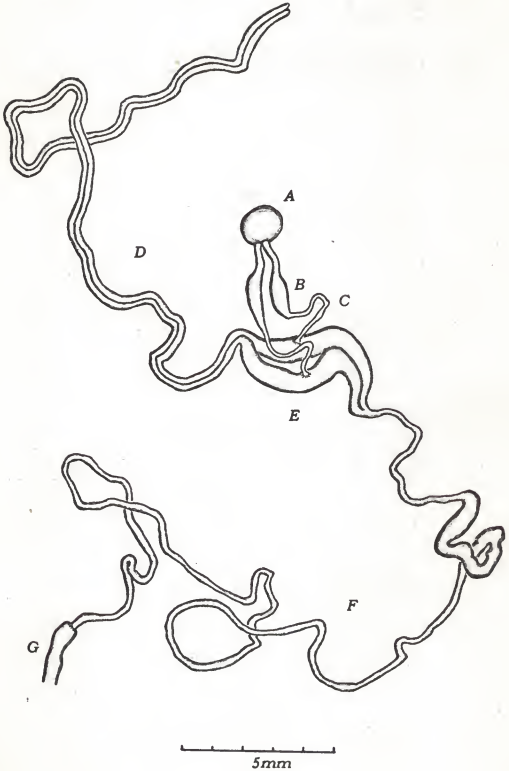

A
D
B
C
3 mm

Figure 56. The male reproductive system:

A. testis
B. seminal vesicle
C. vas deferens
D. accessory gland
E. ductus ejaculatorius duplex
F. ductus ejaculatorius simplex
G. aedeagus


A
B
C
D
E
F
G
5mm

B. Egg

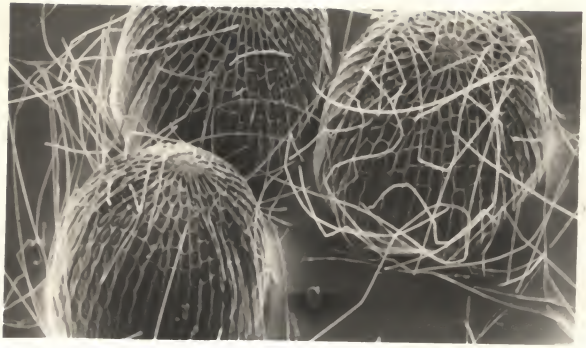
Eggs are oblong spheroid in shape and circular in cross section, with a diameter of approximately o.42 mm. They are greenish when oviposited, turn cream-colored within a few hours and finally become dark just before hatching due to the black color of the larval head. They are laid in clusters of 5 to over 100 in a single layer and are covered with whitish scales from the tip of the female abdomen for the early masses of eggs. Masses of eggs oviposited later are often not covered because the female moth has lost almost all the setae at the abdominal tip (Figure 57). The micropyle, located at the top, is surrounded by a petal-like pattern with the number of petals varying from 7 to 11 (Figure 58). The whole egg is composed of many concave surfaces delineated by higher ridges (Figure 58). Many tiny pores, called aeropyles, can be seen distributed at the edges of the ridges at 1430X (Figure 59). These aeropyles play a principal role in gas and moisture exchange (Hinton, 1969).

C. Larva

There are five larval instars and on occasion six. The first instar larva is about 1 mm long with a whitish body and black head. The body color is highly variable from specimen to specimen in later instars, ranging from light green to olivaceous to near black. The head becomes lighter in color being brownish with a number of near-

Figure 57. Beet armyworm eggs partly covered with scales from tip of female abdomen (X133)

Figure 58. Apex of egg showing micropyle and chorion design (X931)



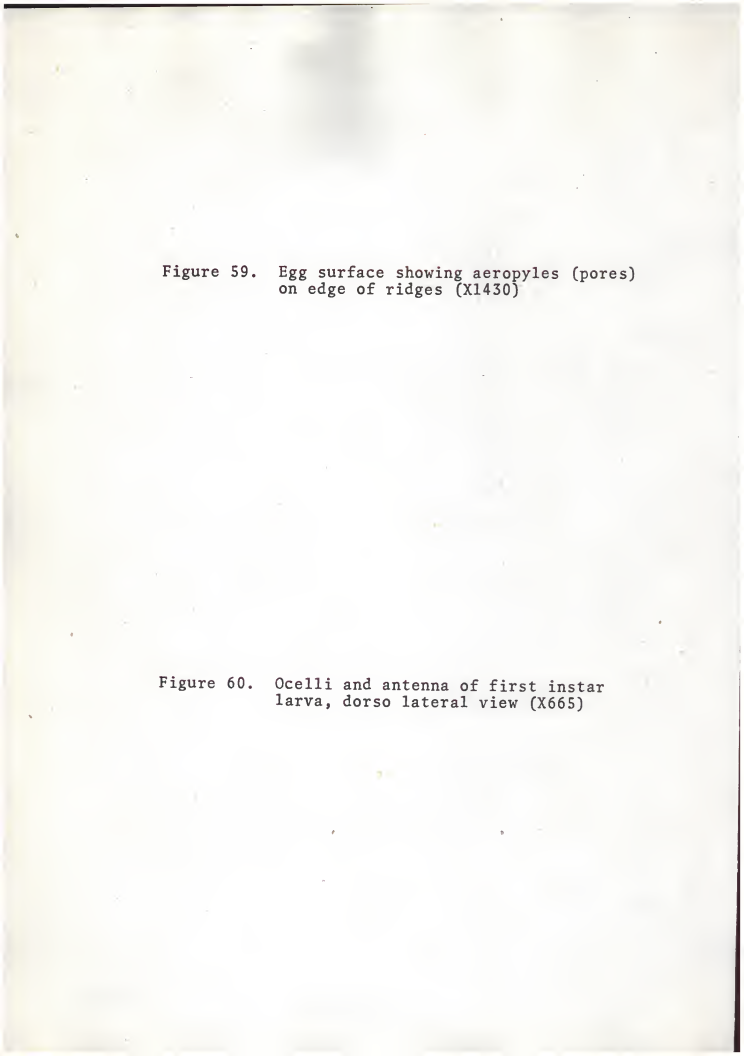
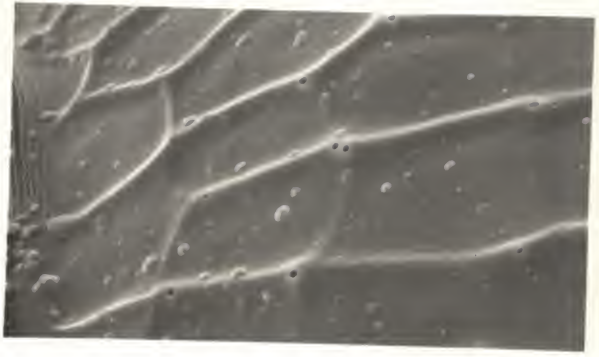
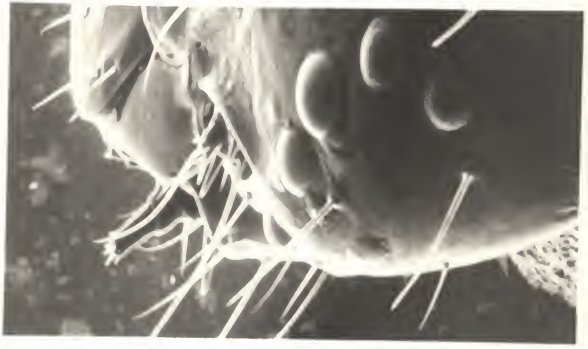


Figure 59. Egg surface showing aeropyles (pores) on edge of ridges (X1430)

Figure 60. Ocelli and antenna of first instar larva, dorso lateral view (X665)





white reticulations on the epicranium. The width of the first instar head capsule is .25 mm (n=60). Full grown larvae measure from 30 to 35 mm.

Only the first instar larva was examined with the scanning electron microscope and its description follows: The 6 ocelli are arranged in a semi-circular position and are unequal in size: the third ocellus is the largest while the first and the sixth ocelli are smaller than the others (Figure 60). Setae on the head are more pointed than those on the body, with lengths varying from 0.02 to 0.08 mm. They are usually curved or bent near the tip while the body setae are more or less straight and thicker. The body setae are also inserted in a raised pinaculum which has a much larger diameter and is navel-like (Figure 61), whereas those on the head have only a simple peritreme (Figure 63). There are numerous tiny spicules on the body evenly distributed almost everywhere except on some transverse folds and the intersegmental positions of the larva (Figure 62).

D. Chromosome Number

Chromosomes are more or less spherical (dot-like), similar to those found in other Noctuids. Definitive chromosome counts with photographic confirmation were obtained from five male larvae and four adults. The haploid chromosome number of the beet armyworm is n=31 (Figure 64, 65). A haploid set of 31 chromosomes is the

Figure 61. Microspines and setae on first instar larva (X1596)

Figure 62. Spiracle of first instar larva (X1410)

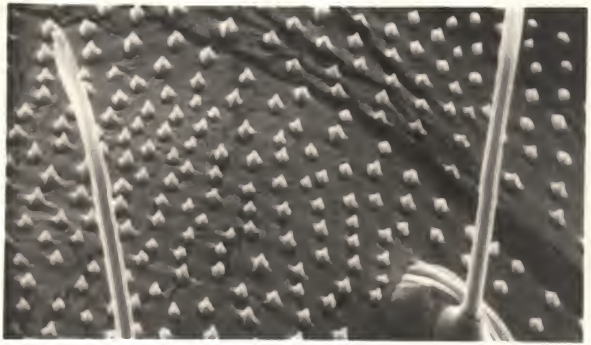


Figure 63. Overall view of anterior portion of first instar larva (X300)

most common number found to date in the Noctuidae (Robinson, 1971). The haploid number of chromosomes varies from 29 to 34 among the 36 Noctuid species studied so far (Robinson, 1971). Another member of the same genus, Spodoptera litura (=Prodenia litura) has the same number (31) of chromosomes as S. exigua (Gupta, 1964).

Sperms and spermatids were observed in the preparation taken from the adults testes (Figure 66, 67). The head of the sperm containing the nucleus looks like the tail but thicker. It has an oblong form followed by a long filamentous tail (Figure 67). The length of the sperms varies from 12.6 to 23.5μ.

Figure 64. Chromosomes of the beet armyworm observed under microscope with phase (n=31) (X1197)

Figure 65. Chromosomes of the beet armyworm observed under microscope without phase (X1197)

Figure 66. Bundles of sperms in testis (X 1197)

Figure 67. Two sperms among nucleus division in testis (X1197)

CHAPTER V

CONCLUSIONS

Based on this research the following morphological distinctions have been determined for the beet armyworm:

1. The female antennae are longer than male antennae.

2. There are six morphologically distinct types of sensilla on the antennae of each sex. The male, however, does have a distinct pattern arrangement of the long sensilla trichodea.

3. Wing areas can be divided into distinct zones based on types of scales.

4. Sexes can be distinguished by the number of frenulum spines on the hindwing. The male has one while the female has three and sometimes four.

5. Scent glands are present in the ventral intersegmental area between the eighth and ninth abdominal segments.

6. The female has two lateral plates on the eighth abdominal segment which serve as the ovipositor. Three types of sensilla are found on these plates.

7. The ninth abdominal segment of the male has a curved uncus and two lateral hooks. These hooks do not occur in all other noctuid genera.

8. Externally, there are many more scales and very few sensilla on the male genitalia compared to the female.

9. The female ovary is composed of four ovarioles. The distinct bursa copulatrix is described as well as the spermatophore.

10. The haploid chromosome number for this species was determined to be n=31.

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BIOGRAPHICAL SKETCH

Ngo Dong was born October 4, 1937, in Hanoi North VietNam. In 1961, he received the B.S. from the University of Hue, South VietNam, and became a teaching assistant in Zoology of the University of Hue. In 1967, he was promoted to Chief of the Zoology Laboratory. In 1968, he became Assistant Dean for Student Affairs in the Faculty of Science. In March 1971, he was sent by the University of Hue to take higher degrees at the University of Florida.

From 1971 until the present time he has pursued work toward the degree of Doctor of Philosophy in Entomology.

On January 11, 1963, Ngo Dong was married to Ton Nu Thanh Chau and his family now includes four children, three boys and one girl.

He is a member of the Newell Entomological Society, the Florida Entomological Society and the Entomological Society of America.

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

Dale H. Habeck, Chairman
Professor of Entomology

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

Harvey L. Cromroy
Professor of Entomology

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

Milledge Murphey
Professor of Entomology

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

Louis C. Kuitert
Professor of Entomology

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

James Soule

James Soule
Professor of Fruit Crops

This dissertation was submitted to the Graduate Faculty of the College of Agriculture and to the Graduate Council, and was accepted as partial fulfillment of the requirements for the degree of Doctor of Philosophy.

June, 1974

Dean, College of Agriculture

Dean, Graduate School